

## **Claims**

1. A method for generating dopaminergic neurons comprising the steps of:
  - (i) providing pluripotent cells;
  - (ii) inhibiting one or more pathway components of a TGF- $\beta$  signaling pathway in said pluripotent cells; and
  - (iii) overexpressing one or more cell fate-inducing polypeptides in said pluripotent cells.
2. The method of claim 1, wherein one of said cell fate-inducing polypeptides is Nurr-1.
3. The method of claim 1, wherein one of said cell fate-inducing polypeptides is PTX3.
4. The method of claim 1, wherein said cell fate-inducing polypeptides are Nurr-1 and PTX3.
5. The method of claim 1, wherein said one or more cell fate-inducing polypeptides is overexpressed by:
  - (i) providing a polynucleotide encoding said cell fate-inducing polypeptide operably linked to a promoter; and
  - (ii) introducing said polynucleotide into said pluripotent cells under conditions suitable for expression of said polynucleotide.
6. The method of claim 1, wherein said pluripotent cells are human pluripotent cells.

7. The method of claim 1, wherein said pluripotent cells are mouse, rat, porcine, or non-human primate pluripotent cells.
8. The method of claim 6, wherein said pluripotent cells are embryonic stem cells.
9. The method of claim 1, wherein said TGF- $\beta$  signaling pathway is the Nodal signaling pathway.
10. The method of claim 1, wherein said TGF- $\beta$  signaling pathway is the Activin signaling pathway.
11. The method of claim 1, wherein said TGF- $\beta$  signaling pathway is the BMP2, BMP4, or BMP7 signaling pathway.
12. The method of claim 1, wherein said TGF- $\beta$  signaling pathway component is selected from the group consisting of Nodal, Cryptic, Cripto, Activin, Activin receptor I, Activin receptor II, Activin receptor IIb, TGF- $\beta$  receptor, ALK-1, ALK-2, ALK-3, ALK-4, ALK-6, ALK-7, BMP2, BMP4, BMP7, BMPRIa, BMPRIb, BMPRII, Smad2, Smad3, Smad4, Smad5, and Smad6.
13. The method of claim 1, wherein said TGF- $\beta$  signaling pathway component is Smad4.
14. The method of claim 1, wherein said TGF- $\beta$  signaling pathway component is Cripto.

15. The method of claims 1, wherein said dopaminergic neurons are A9 dopaminergic neurons.

16. The method of claim 1, wherein said pathway component is inhibited by gene knockout of the nucleic acid encoding said component.

17. The method of claim 1, wherein said pathway component is inhibited by overexpressing small interfering RNA complementary to the mRNA encoding said component in said pluripotent cells.

18. The method of claim 1, wherein said pathway component is inhibited by overexpressing antisense oligonucleotide of the nucleic acid encoding said component in said pluripotent cells.

19. The method of claim 1, wherein said pathway component is inhibited by contacting said pluripotent cells with antibodies that specifically bind to said pathway component.

20. The method of claim 1, wherein said pathway component is inhibited by overexpressing a dominant negative version of said pathway component in said pluripotent cells.

21. A method for treating a neurodegenerative disease in a patient, said method comprising the steps of:

(i) providing dopaminergic neurons generated by a method comprising the steps of:

(a) providing pluripotent cells,

- (b) inhibiting one or more pathway components of a TGF- $\beta$  signaling pathway in said pluripotent cells, and
  - (c) overexpressing one or more cell fate-inducing polypeptides in said pluripotent cells; and
- (ii) transplanting said dopaminergic neurons into the brain of said patient.

22. The method of claim 21, wherein said neurodegenerative disease is Parkinson's disease.

23. The method of claim 22, wherein said dopaminergic neurons are transplanted into the caudate, the putamen, or the substantia nigra of said patient.

24. The method of claim 21, wherein one of said cell fate-inducing polypeptides is Nurr-1.

25. The method of claim 21, wherein one of said cell fate-inducing polypeptides is PTX3.

26. The method of claim 21, wherein said cell fate-inducing polypeptides are Nurr-1 and PTX3.

27. The method of claim 21, wherein said one or more cell fate-inducing polypeptides is overexpressed by:

- (i) providing a polynucleotide encoding said cell fate-inducing polypeptide operably linked to a promoter; and
- (ii) introducing said polynucleotide into said pluripotent cells under conditions suitable for expression of said polynucleotide.

28. The method of claim 21, wherein said pluripotent cells are human pluripotent cells.
29. The method of claim 21, wherein said pluripotent cells are mouse, rat, porcine, or non-human primate pluripotent cells.
30. The method of claim 21, wherein said pluripotent cells are embryonic stem cells.
31. The method of claim 21, wherein said TGF- $\beta$  signaling pathway is the Nodal signaling pathway.
32. The method of claim 21, wherein said TGF- $\beta$  signaling pathway is the Activin signaling pathway.
33. The method of claim 21, wherein said TGF- $\beta$  signaling pathway is the BMP2, BMP4, or BMP7 signaling pathway.
34. The method of claim 21, wherein said TGF- $\beta$  signaling pathway component is selected from the group consisting of Nodal, Cryptic, Cripto, Activin, Activin receptor I, Activin receptor II, Activin receptor IIb, TGF- $\beta$  receptor, ALK-1, ALK-2, ALK-3, ALK-4, ALK-6, ALK-7, BMP2, BMP4, BMP7, BMPRIa, BMPRIb, BMPRII, Smad2, Smad3, Smad4, Smad5, and Smad6.
35. The method of claim 21, wherein said TGF- $\beta$  signaling pathway component is Smad4.

36. The method of claim 21, wherein said TGF- $\beta$  signaling pathway component is Cripto.
37. The method of claims 21, wherein said dopaminergic neurons are A9 dopaminergic neurons.
38. The method of claim 21, wherein said pathway component is inhibited by gene knockout of the nucleic acid encoding said component.
39. An isolated mammalian pluripotent cell expressing a recombinant cell fate-inducing polypeptide and having a functional disruption of a TGF- $\beta$  signaling pathway component.
40. The cell of claim 39, wherein said cell is a human cell.
41. The cell of claim 39, wherein said cell fate-inducing polypeptide is Nurr-1 or PTX-3.
42. The cell of claim 39, wherein said functional disruption is a result of a homozygous deletion of a gene encoding a TGF- $\beta$  signaling pathway component.
43. The cell of claim 39, wherein said functional disruption is a result of a missense mutation in a gene encoding TGF- $\beta$  signaling pathway component.
44. The cell of claim 39, wherein said TGF- $\beta$  signaling pathway component is selected from the group consisting of Nodal, Cryptic, Cripto,

Activin, Activin receptor I, Activin receptor II, Activin receptor IIb, TGF- $\beta$  receptor, ALK-1, ALK-2, ALK-3, ALK-4, ALK-6, ALK-7, BMP2, BMP4, BMP7, BMPRIa, BMPRIb, BMPRII, Smad2, Smad3, Smad4, Smad5, and Smad6.

45. The cell of claim 39, wherein said TGF- $\beta$  signaling pathway component is Smad4.

46. The cell of claim 39, wherein said TGF- $\beta$  signaling pathway component is Cripto.